MEMORY: WHERE AND HOW MEMORIES STORED?

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ABSTRACT

Human brain is considered to be the most complex structure in the universe. Research in memory began in early of nineteenth century. After more than forty years of study, they found where memory is stored temporarily, i.e. in the hippocampal formation and adjacent, anatomically related cortex: the perirhinal and parahippocampal cortices. Serotonin acts as neurotransmitter in implicit short-term memory storage, while glutamate involves in explicit short-term memory storage. For long-term memory storage, both implicit and explicit used the same core-signaling pathway, PKA, MAPK, and CREB-1. Short-term memory storage, either implicit or explicit does not required new protein synthesis, but undergone covalent modification of preexisting protein which cause increase synaptic strength, while the long term memory storage involve activation of gene expression, new protein synthesis and the formation of new connection.

***Keywords**: memory, implicit, explicit, hippocampus*.

INTRODUCTION

Human brain is considered the most complex structure in the universe (Woolley, 2001). The brain weighs about 3 pounds, comprises about 97% of the central nervous system, 2% of body weight, and consumes 20% of body's energy. It is estimated that the brain is a collection of some 30 to 100 billion neurons with one trillion connections (Greengard, 2001; Anonymous1, 2003).

Hermann Ebbinghans performed the first human simple experimental method for studying learning and memory in 1885, followed a few years later in experimental animals by Ivan Pavlov and Edgar Thorndike. This experimental method of studying learning and memory lead to the development of empirical school of psychology called behaviorism. Behaviorists concentrated on examining objectively and precisely the relationship between specific physical stimuli and observable responses in intact animal, but largely ignored mental processes. By the 1960's, emerges new discipline of science named cognitive psychology. Unlike behaviorists, it was also concerned on the flow of sensory receptors to its eventual use in memory and action. The neuroscience has grown rapidly over the last half century. This has provided a new framework for the study of memory, perception, action, language, and conscious awareness. Cognitive neuroscience emerges as fusion of two disciplines, psychology and neurobiology. The fusion of these two disciplines was facilitated as well by the emergence of coherent neuroscience. In the cognitive neuroscience, an interdisciplinary approach to the nervous system might be usefully applied to the analysis of cognition (Miller, 1998).

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Within the discipline of cognitive neuroscience this paper is written focusing on memory and learning, as introduction, especially on where, and how in the molecular level memory is stored. Very important definition must be introduced here; firstly, implicit memory – a memory for precept and motor skills – involves a variety of anatomical systems (Schacter, 1994). Implicit memory is expressed through performance, without conscious recall of past episode. For example, one form of implicit memory, that for conditioned fear, involves the amygdale. Secondly, explicit memory (declarative memory) – a memory for facts, places, and events – requires the hippocampus and related medial temporal lobe structures. Explicit memory requires conscious recall (Kandels, 2001).

WHERE ARE MEMORIES STORED?

At the beginning of the 19th century, F.J. Gall studied the surface of the skulls of individuals, and divided the brain underneath into at least 27 regions. Each region corresponds to a specific mental faculty. He thought that even the most abstract and complex of human traits, such as generosity and secretiveness, are localized to discrete areas of the brain. Gall called this anatomically oriented approach to personality organology. Later it was evident that he misidentified the function of most parts of the cortex.

P.Flourens (1820) subjected Gall's ideas into experimental analysis. From this experiments Flourens concluded that individual site in the brain are not sufficient for specific behaviors such as sexual behavior and romantic love and that all regions of the brain especially the cerebral hemispheres of the fore brain participate in every mental function. He proposed that any part of the cerebral hemisphere is able to perform all the functions of the hemisphere.

In the period from 1920 to 1950, the debate between cortical localization and equipotentiality in cognitive function dominated thinking about mental process, including memory. Lashley (1929) explored the cerebral cortex in the rat, systematically removing different cortical areas. He found that there was no particular any brain region involved specifically in memory storage. Hebb (1949) in his book the organization of behaviors suggested that memory storage involves many parts of the brain. There was no apparent single memory center exists, and many parts of the nervous system participate in the representation of any single event. B. Milner (1957) was Hebb students who described the remarkable patient H.M. H.M. had sustained a bilateral resection of the medial structures of the temporal lobe in 1953 to relieve severe epilepsy. Following the surgery H.M. had a very profound impairment of recent memory in the apparent absence of other intellectual loss. The works of W. Penfield in association with Milner from 1955 until 1964 on their patients and further proven by an autopsy finally showed that when both side of hippocampus were deprived the patient suffered a severe, persistent, and generalized impairment of recent memory.

In the year of 1991, Mort Mishkin and Zola Morgan established an animal model of human amnesia in the monkey. With the model, the question of precisely which structures within the medial temporal lobe were important could be systematically explored. The important structures are the hippocampus proper, the dentate gyres, the subicular complex, and the enthorhinal cortex (which together comprise the hippocampal formation) and adjacent, anatomically related cortex: the perirhinal and parahippocampal cortices.

A key feature of medial temporal lobe function is that the medial temporal lobe is involved in memory for a limited period after learning. The medial temporal lobe structures direct a gradual process of organization of cortical representations, for examples, by gradually binding together the multiple, geographically separate cortical regions that together store memory for a whole event. After sufficient time has passed, the hippocampal formation is not needed to support storage or retrieval of declarative memory and long-term memory is fully dependent on neocortex.

The different components of the medial temporal lobe need not have equivalent roles in declarative memory; different structures within the medial temporal lobe are likely to carry out different sub functions. As damage increase, fewer strategies may be available for storing memory, with the result that memory impairment become more severe.

LEARNING AND MEMORY

Learning is the process by which we acquire new knowledge, whereas memory is the process by which we retain that knowledge over time. Memory is the out come of learning. It is not possible to consider learning without memory or conversely, memory without learning (Anonymous 2).

In Aplysia, simple reflex could be modified by three different form of learning: habituation, sensitization, and conditioning. Memory storage for each type of learning in Aplysia has two phases: a transient memory that lasts minutes and enduring memory that last days (other term used are short-term and long-term memory). Conversion of short term to long-term memory storage requires spaced repetition (Kandel, 2001).

The simple reflex in Aplysia is in fact a simple learning behavior and is called implicit learning, as a result acquires an implicit memory or non-declarative memory. Explicit (declarative) learning/memory involves knowledge about people, facts, places, and events. Many studies in mice confined to memory for space, a complex form of explicit memory storage. There are at least two theories proposed on how memory stored. First, memory is stored in the growth of new connections. Second, memory is stored dynamically by self-reexciting chain of neuron.

To explore the molecular basic of learning and memory is in fact to examine the change in the area close to synapses, either pre or postsynaptic area. Synapses can convert electrical impulse into chemical signals and back again, as well as modulate the strength of the transmitted signals. This ability to modify the strength of transmission known as synaptic plasticity is thought to be the cellular basis of the brain's ability to compute, learn, and remember (Dorunz, 2002).

IMPLICIT LEARNING AND MEMORY

E.R. Kandel (2001) searches the molecular basic of learning and memory in Aplysia by focusing initially on one type of learning i.e. sensitization. It is a form of learning of fear where by Aplysia on receiving an aversive shock to a part of the body such as the tail, it recognizes the stimulus as aversive and learns to enhance its defensive reflex responses to variety of subsequent stimulus applied to the siphon, even innocuous stimulus. A single shock gives rise to a memory last only minutes; this short-term memory does not require the synthesis of new protein. In contrast, four or five spaced shock to the tail give rise to a memory lasting several days, the long-

term memory does required the synthesis of new protein.

Molecular Mechanisms

a. Short-term sensitization (Figure 1A)

Stimulation of sensory neurons in the tail activates specific interneurons that facilitate the sensitization. Synapses of the facilitating interneurons form axo-axosynaptic contacts with axons of the sensory neuron. Serotonin released by the facilitating interneurons binds to two types of G protein (Gs and Go)-coupled receptors, leading to respective activation of cAMP-dependent protein kinase (protein kinase A, PKA) and protein kinase C. Activation of Gs (s = stimulatory)-coupled receptors on the sensory axons increases axonal cyclic AMP levels. The elevated cAMP activates cAMP-dependent protein kinase (PKA), which then phosphorylates K^+ channels in the sensory axon. Phosphorylation inactivates the hyperpolarizing K^+ channels in the sensory axon, leading to prolonged action potential and increased duration of Ca^{2+} influx through voltage-sensitive Ca^{2+} channels. On the other hand, activation of Go (o = other)-coupled receptors activates phospholipase C (PLC) that catalyzes the formation of diacylglycerol (DAG), a PKC activator. PKC, together with PKA, phosphorylates L-type Ca^{2+} channels, resulting in opening of the channels. The net effect is that more Ca^{2+} flows into the axon, leading to greater transmitter release and increased gill withdrawal.

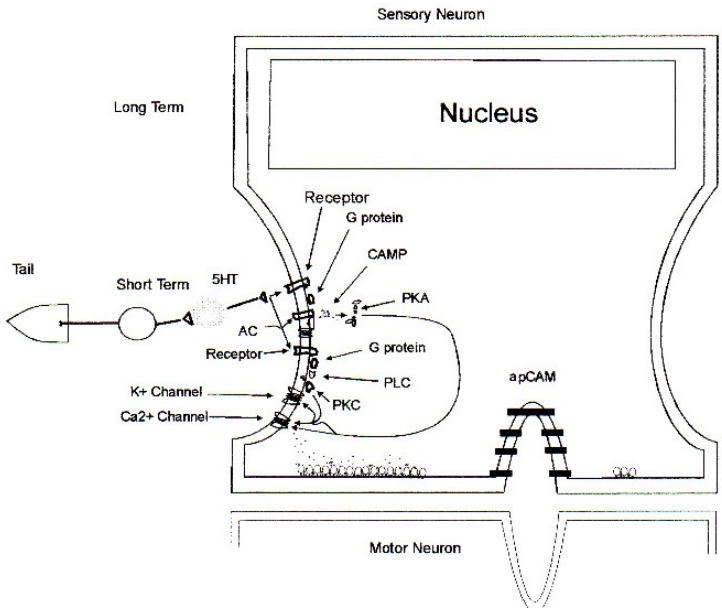

Figure 1A. Effect of Short sensitization on the monosynaptic component of the gill withdrawal reflex of Aplasia (Adapted from: Albert, 1989; Anonymous 2, 2002; Kandel 2001)

b. Long-term sensitization (Figure 1B)

Repeated stimulation increases the levels of cAMP (and thus PKA) to rise and persist for several minutes. Activated PKA then recruits the mitogen-activated protein kinase (MAPK) and together they translocate to the nucleus. In the nucleus, PKA phosphorylates and activates the transcription factor CREB1 (cAMP-response element-binding protein), whereas MAPK phosphorylates and remove the repressive action of CREB-2, an inhibitor of CREB-1. Then, CREB-1 activates several immediate-response genes encoding functionally important proteins, including:

1. Ubiquitin hydrolase: a component of a specific ubiquitin protease that leads to the proteolysis of the regulatory subunit of PKA, resulting in *persistent activity of PKA, leading to persistent phosphorylation of the substrate proteins of PKA, including both CREB-1 and the protein involved in the short-term process. (*Note: This is the simplest mechanism for long-term memory**.** That is, PKA,

critical for the short-term process, is made persistently active for days by repeated training, without requiring a continuous signal. The kinase becomes autonomous, and does not require serotonin, cAMP or PKA).

2. C/EBP (CAAT Enhancer Binding Protein): transcription factor that binds to the DNA response element CAAT, which activates genes that encode proteins important for the growth of new synaptic connections. (Kandel, 2001; Anonymous 2, 2002).

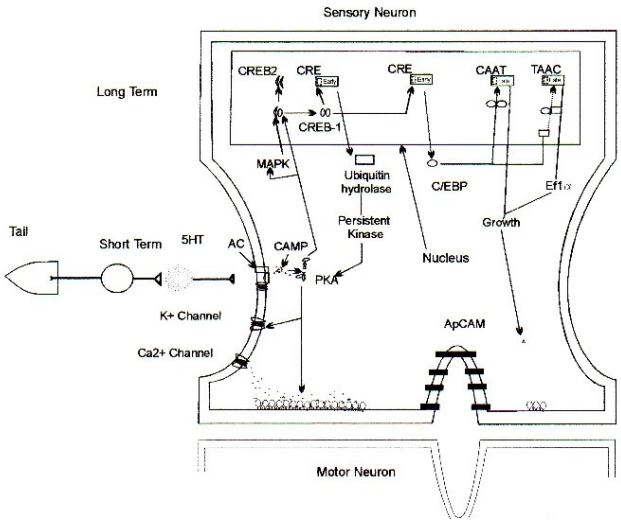

Figure 1B. Effect of Short and Long Term sensitization on the monosynaptic component of the gill-withdrawal reflex of Aplasia (Redrawn from Kandel, 2001).

EXPLICIT LEARNING AND MEMORY

Explicit memory unlike implicit memory requires conscious recall and concerned with memory for people, places, and event. Explicit memory involve in specialized anatomical system in the medial temporal lobes and structure deep to it, the hippocampus. The hippocampus contains a cellular representation of extra-personal space, a cognitive map of space, and lesion of hippocampus interfere with spatial task. More over, within the hippocampus the perforant path, a major path way exhibits activity dependent plasticity, a change now called long-term potentiation (LTP) (Kandel, 2001).

LTP of synaptic transmission in the hippocampus is the leading experimental model for the synaptic change that may underlie learning and memory (Malenka, 1999; Wooley, 2001). How ever LTP occur not only in hippocampus, and it function probably not only related to memory stored, but it is a fundamental property of the majority of exitory synapses in the mammalian brain, and as such, is likely to serve many function. The experimental design at present mostly carried out in rat and recorded in the hippocampal CA1 region following stimulation of CA3 Schaffer collateral (Sch) (Figure 3A).

Early phase LTP obtained with a single train of stimuli given for one second at 100 Hz. Early LTP lasts 2-3 hours.Late phase of LTP occurs with four trains of stimuli separated by 10 min. The late LTP lasts 24 or more hours (Figure 3B).

Molecular Mechanisms

a. Early phase LTP (E-LTP) (Figure 2)

A single train of high-frequency tetanus stimulation results in: (1) an increase Ca^{2+}-dependent exocytosis of glutamate presynapticly. In postsynaptic neuron it will (2) increase activation of AMPA (α amino 3 hydroxy 5 methyl 4 isoxazole propionic acid) receptors result in (3) increase depolarization that relieves Mg^{2+} blockage of the NMDA (N Methyl D Aspartate) receptors channel, allowing Ca^{2+} entry into the postsynaptic cell. Stimulation also (4) activates metabo-tropic glutamate receptors (mGluR), which can cause phosphorylation of NMDAR, results in further increase in intracellular Ca^{2+} level. In addition it (5) activate voltage dependent Ca^{2+}

channels, and increase Ca^{2+} intra cellular even further. The rise in Ca^{2+} triggers Ca^{2+}-dependent kinases (Ca^{2+}/calmodulin dependent kinase, CK and PKC) as well as the protein tyrosine kinase, Fyn that together induces LTP. Some suggestion of (6) activation of nNOS, result in release of NO, a retrograde messenger that increase synaptic transmission. (Kandel, 2001; Anonymous 2, 2002; Duty, 2002.)

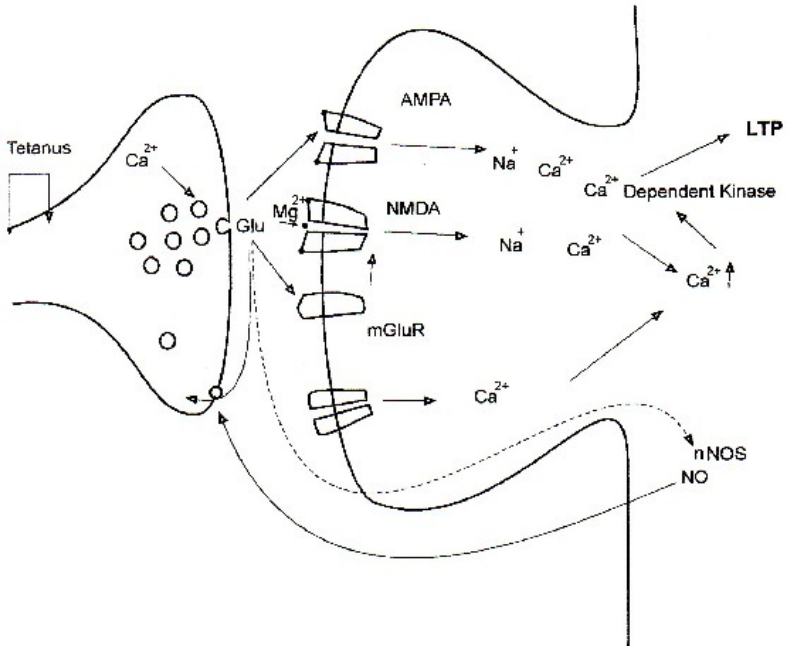

Figure 2. Cellular Basis of LTP Induction (Adapted from Anonymous 2, 2002: Duty, 2002)

b. Late phase LTP (L-LTP) (Figure 3 A B C)

With repeated trains of stimuli, the Ca^{2+} influx also recruits an adenylyl cyclase, leading to the activation of PKA that plays a critical role in the transformation of short-term explicit memory into long-term memory. L-LTP, like long-term storage of implicit memory, requires pathways involving PKA, MAPK and CREB. PKA and MAPK are transported to the nucleus to activate CREB, which, in turn, activates effectors for growth (tPA= tissue plasminogen activator, BDNF= brain-derived neurotrophic factor) and regulators (C/EBPβ=CAAT Enhancer Binding Protein), resulting in the synthesis of new proteins and structural changes, including the formation of new connections.

The balance between protein phosphorylation governed by PKA and dephosphorylation determines the threshold for hippocampal synaptic plasticity and memory storage. It has been proven that endogenous Ca^{2+}-sensitive phosphatase calcineurin acts as a constraint on this balance. It might be worthwhile to mention here that other modulator inputs, such as dopamine signaling pathway, may also regulate the adenylyl cyclase activity, thereby participating in the processes of long-term memory. However, the detail mechanism of it still to be describes (Greengard, 2001).

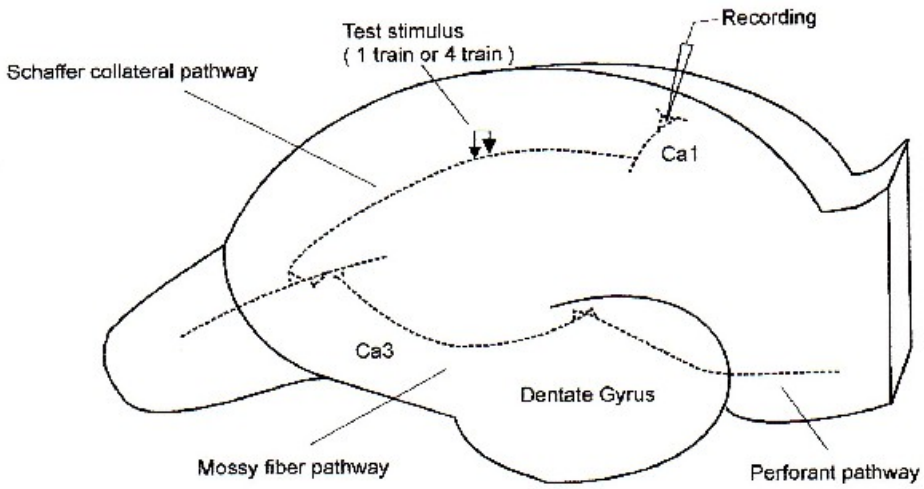

Figure 3A. Three major pathway, each of which give rise to LTP

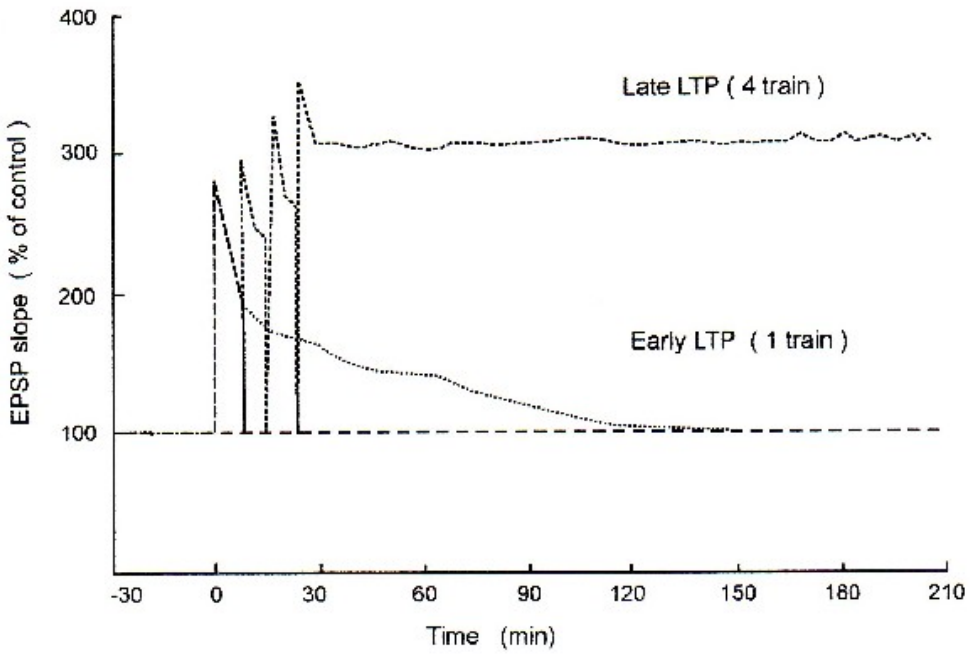

Figure 3B. The early and late phase of LTP in the Schaffer collateral. Early: One second at 100 Hz (last 2 hours). Late: Four trains at 10-minute interval (last > 24 hours).

(Redrawn from Kandel, 2001)

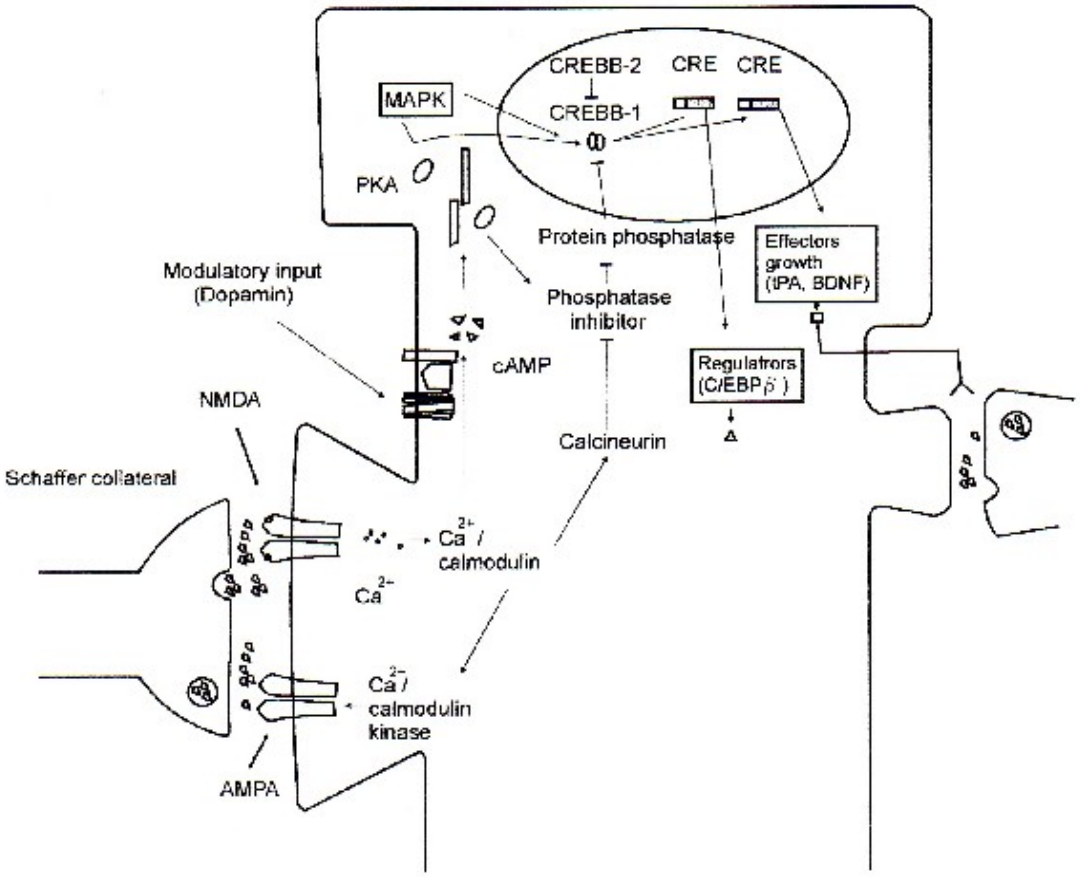

Figure 3C. A model for late LTP in the Schaffer collateral pathway
(Redrawn fro Kandel 2001)

CONCLUDING REMARKS

The study so far led to the conclusion that memory storage involves in the synaptic changes. The short term memory result in covalent modification of preexisting protein, which cause increase synaptic strength, while the long term memory storage involve activation of gene expression, new protein synthesis and the formation of new connection. The study of memory will continue for many years to come, since there are so many questions seeking the answers. For instances, how do different parts of the brain interact. Why hippocampus needed only for short period in memory storage. What is the molecular mechanism in memory recall, and many others formidable task to be encountered.

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